

# Defoliation and Fruit Coloration in 'Fuji'/M.9 Apple Affected by Cu-EDTA and Fe-EDTA Foliar Spray

Heon-Kyu Lim<sup>1†</sup>, Hyunsuk Shin<sup>2†</sup>, In-Chang Son<sup>3</sup>, Youngjae Oh<sup>1,4</sup>, Keumsun Kim<sup>1</sup>, Hyeondae Han<sup>1</sup>, Sewon Oh<sup>1</sup>, and Daeil Kim<sup>1\*</sup>

<sup>1</sup>Department of Horticultural Science, Chungbuk National University, Cheongju 28644, Korea

<sup>2</sup>Department of Horticultural Science, Gyeongnam National University of Science & Technology, Jinju 52725, Korea

<sup>3</sup>Planning and Coordination Division, National Institute of Horticultural and Herbal Science, Rural Development Administration, Wanju 55365, Korea

<sup>4</sup>Department of Horticultural Science, IFAS Gulf Coast Research and Education Center, University of Florida, 14625 County Road 672, Wimucama, FL 33598, USA

\*Corresponding author: [dkpomo@cbnu.ac.kr](mailto:dkpomo@cbnu.ac.kr)

†These authors contributed equally to this work.

## Abstract

The objective of this study was to determine whether two chelate compounds (Cu-EDTA and Fe-EDTA) applied as a foliar spray could effectively defoliate 'Fuji'/M.9 apple trees and improve fruit quality and fruit coloration. Foliar spray with these two chelate compounds at 30 days before harvest significantly defoliated fruit cluster leaves, which improved light interception of 'Fuji'/M.9 apple trees. However, Cu-EDTA non-selectively defoliated bourse shoot leaves and true leaves, which are involved in photosynthesis during the late period of fruit growth. Cu-EDTA markedly reduced fruit weight, soluble solids content, and fruit coloration compared to Fe-EDTA. Regarding Fe-EDTA concentration, 3% Fe-EDTA was more effective at enhancing fruit coloration but lowered fruit weight compared to 1% Fe-EDTA. Consequently, we found that 1% Fe-EDTA induced more selective defoliation of fruit cluster leaves than Cu-EDTA and was more effective in improving fruit quality and fruit coloration of 'Fuji' apple trees.

**Additional key words:** abscission, anthocyanin, chemical defoliation, coloration, leaf

## Introduction

'Fuji' apple occupies 80% of the apple growing area in Korea. This cultivar has several advantages including high quality and long shelf life. However, its fruit color has been hindered by inadequate light interception in the tree, temperature, and nitrogen and soluble solids contents (Vestheim, 1970; Choi et al., 2000). In order to improve fruit color, an ideal tree shape and adequate fertilization are required. Most growers try to improve fruit color using management practices such as hand defoliation, removal of water sprouts, training, and reflective film mulching to increase light interception (Jackson et al., 1971; Choi et al., 2000). However, these methods are labor-intensive, which can increase the cost of production. Therefore, it is necessary to develop defoliant that can

Received: February 7, 2019

Revised: April 23, 2019

Accepted: May 3, 2019

 OPEN ACCESS



HORTICULTURAL SCIENCE and TECHNOLOGY  
37(4):448-454, 2019  
URL: <http://www.kjhst.org>

pISSN : 1226-8763  
eISSN : 2465-8588

This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

Copyright©2019 Korean Society for Horticultural Science.

This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry (IPET) through Agri-Bio Industry Technology Development Program (or Project), Funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (315003-5).

reduce labor (Yim et al., 2000).

In previous studies on defoliant, 2,3,5-triiodobenzoic acid (TIBA), an anti-auxin growth regulator, was used to improve fruit coloration in 'Fuji' and 'Jonathan' apples (Kim and Yun, 1971). However, it was banned due to food safety concerns. Later, abscisic acid (ABA) and its synthetic intermediate STC-4771 were applied to 'Fuji' and 'Jonathan' apples for defoliation. However, their defoliation effect was inconsistent (Kamuro and Sakai, 1995; Yim et al., 2000). The chelate compounds Cu-EDTA and Fe-EDTA are very effective defoliant and are being used as a foliar spray to advance the period of digging and to protect apple trees from freezing injury on apple nursery stocks in autumn (Knight, 1983; Faby, 1988).

The present study was undertaken to investigate the effectiveness of two types of chelate compounds, Cu-EDTA and Fe-EDTA, as an individual or a mixed foliar spray, on defoliation and fruit coloration of 'Fuji'/M.9 apple.

## Materials and Methods

### Plant Material and Treatment of Chelate Compounds

Eight-year-old 'Fuji'/M.9 apple trees cultivated at Chungbuk Agricultural Research and Extension Services in Cheongju, Korea were used in this study. Cu-EDTA (Ethylenediaminetetraacetic acid copper disodium salt tetrahydrate, Junsei Chemical Co., Ltd, Japan) and Fe-EDTA (EDTA ferric sodium salt trihydrate, 98.0%, Samchun Pure Chemical Co., Korea) chelate compounds were used as a foliar spray. The control (sprayed with distilled water) and treatments were as follows: (1) 1% Cu-EDTA, (2) 3% Cu-EDTA, (3) 1% Fe-EDTA, (4) 3% Fe-EDTA, and (5) 1% Cu-EDTA + 1% Fe-EDTA. A total of 18 trees with similar vigor and fruiting were selected (3 trees per treatment). Only one main branch of each tree was used for each treatment. These trees were sprayed on September 29<sup>th</sup>, 2009 at 30 days before harvest. During spraying, trees were isolated from each other using waterproof cloth to prevent contamination from other treatment solutions.

### Changes in the Chelate Compound-Treated Leaves

Leaves were classified into fruit cluster leaves, bourse shoot leaves, and true leaves. The defoliation rate was measured every 2 days after the treatments. The photosynthetic rate was measured with a photosynthesis analyzer (LC pro+, ADC Bio Scientific Ltd., Hoddesdon, Herts, UK) using three leaves with similar leaf size selected from each tree. The light-emitting diode (LED) light intensity of the photosynthesis analyzer was adjusted to a photosynthetic photon flux density (PPFD) of  $900 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  and the temperature of the measuring block was adjusted to be equal to field temperature. Leaf chlorophyll content was determined using a chlorophyll meter (SPAD-502, Minolta, Japan).

### Characteristics of Harvested Fruit

Characteristics of harvested fruit were investigated on October 30<sup>th</sup>, 2009. Ten fruit was sampled randomly from each main branch. Fruit coloration was determined by measuring three spots of the stem end of each apple using a chromatograph (CR-300, Minolta, Japan). Firmness of the peel and flesh was measured using a durometer (FHM-5, Takemura, Japan) with a  $\text{Ø}5 \text{ mm}^2$  firmness-needle. Soluble solids content was measured with a digital refractometer (Digital refractometer PR-101, Atago, Japan) using juice obtained by crushing and squeezing the flesh. Acidity was

analyzed by squeezing the flesh, filtering the juice, adding 20 mL of distilled water to 5 mL of the juice, and titrating the mixture with 0.1 N NaOH. Data were obtained by converting the amount of NaOH consumed until the discoloring point to the amount of malic acid. Anthocyanin content was assessed by collecting 10 peel discs using a cork borer ( $\text{\O}1.1$  cm), dipping them in 20 mL of anthocyanin extraction liquid [MeOH (85):0.1 N HCl (15)] in darkness at 4°C for 24 h, and measuring with a 530 nm UV/visible spectrophotometer (Ultrospec 4000, Pharmacia Biotech, Sweden).

### Statistical Analysis

Statistical differences were assessed via analysis of variance (ANOVA) with the SAS 9.4 software package (SAS Institute Inc., Cary, NC, USA). Mean differences of data were established by Duncan's multiple range tests.

### Results and Discussion

In this study, leaves of investigated 'Fuji'/M.9 apple trees were classified into fruit cluster leaves, bourse shoot leaves, and true leaves. After treatment with the two chelate compounds, the defoliation rate of 'Fuji'/M.9 apple trees increased significantly compared to that of the control for all types of leaves (Fig. 1). Particularly, Cu-EDTA treatments resulted in

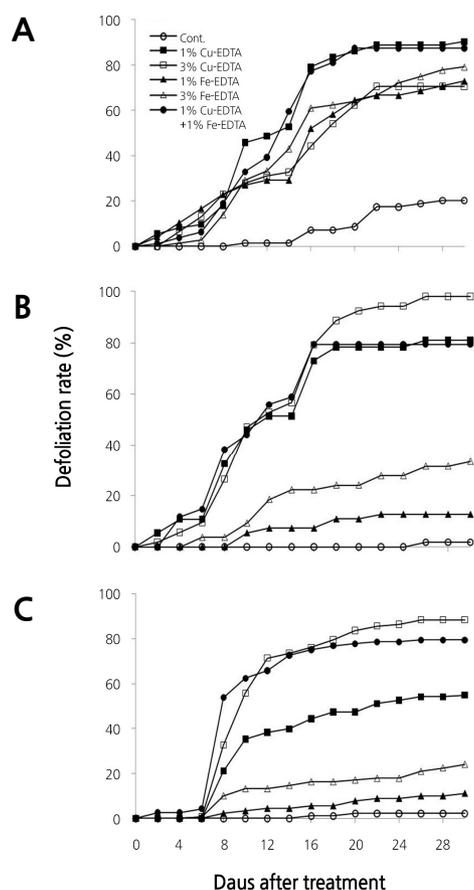


Fig. 1. Defoliation rates of 8-year-old 'Fuji'/M.9 apple trees after the application of a Cu-EDTA and/or Fe-EDTA foliar spray. A, Fruit cluster leaves; B, Bourse shoot leaves; C, True leaves.

higher defoliation rates than Fe-EDTA treatments regardless of leaf type. Ben-Yehoshua and Biggs (1970) reported that Cu-EDTA can produce a larger amount of ethylene than Fe-EDTA, which induces more defoliation. Regarding the effect on leaf type, Cu-EDTA markedly defoliated all types of leaves and the mixed treatment of 1% Cu-EDTA with 1% Fe-EDTA also defoliated all types of leaves, which was similar to the effect of the individual Cu-EDTA treatments. Fe-EDTA remarkably defoliated fruit cluster leaves (Fig. 1A). However, it had less effect on bourse shoot leaves and true leaves (Fig. 1B, C). In addition, the photosynthetic rate of true leaves was the highest in the 1% Fe-EDTA treatment but the lowest in the 3% Cu-EDTA treatment (Fig. 2). Photosynthetic rates in other treatments were in the mid-range. Leaves assimilate photosynthates (Taiz and Zeiger, 2002), and excessive defoliation can lead to a reduced photosynthetic rate for fruit enlargement, reduced soluble solids content, and reduced fruit coloration (AAES, 1985; Srisook et al., 2016; Matsumoto et al., 2017). Therefore, to improve fruit quality, it is better to remove mainly fruit cluster leaves which have a relatively lower photosynthetic rate compared to leaves in other positions (Choi et al., 2000). For this reason, a foliar spray with Fe-EDTA is considered to be better as a defoliant for 'Fuji'/M.9 apple trees.

Fig. 3 shows changes of the specific color difference sensor value (SCDSV) measured by the chlorophyll meter in true leaves at 16 days after the Cu-EDTA and/or Fe-EDTA treatments when the defoliation rate reached almost the maximum in all treatments. None of the chelate treatments significantly decreased the leaf chlorophyll content compared to the

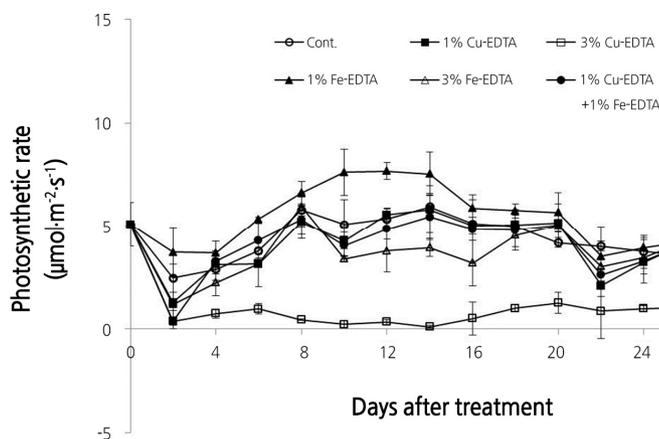


Fig. 2. Photosynthetic rates of true leaves of 8-year-old 'Fuji'/M.9 apple trees after the application of a Cu-EDTA and/or Fe-EDTA foliar spray. Bars represent standard error of means from three replications.

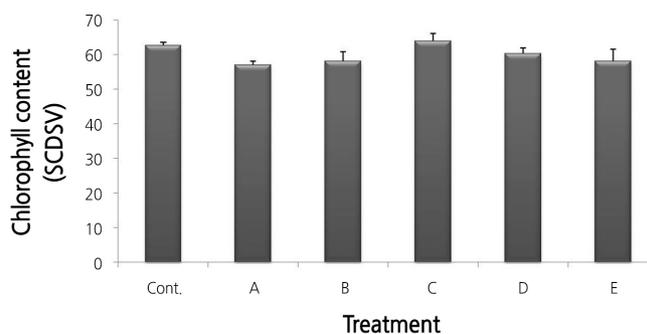


Fig. 3. Chlorophyll contents of true leaves of 8-year-old 'Fuji'/M.9 apple trees at 16 days after the application of a Cu-EDTA and/or Fe-EDTA foliar spray. Cont., water spray; A, 1% Cu-EDTA; B, 3% Cu-EDTA; C, 1% Fe-EDTA; D, 3% Fe-EDTA; E, 1% Cu-EDTA + 1% Fe-EDTA. Bars represent standard error of means from 15 replications.

control. In deciduous fruit trees, nitrogen and photosynthates in leaves should be transferred to stems of the trees before leaf shedding for regrowth in the following year (Millard, 1996; Furze et al., 2019). In this study, the foliar spray with chelate compounds resulted in defoliation without any significant change in leaf chlorophyll content in comparison with the control. This suggests that defoliation might have happened rapidly without the natural process of leaf senescence. Consequently, the transfer of nitrogen and photosynthates from leaves to the stems might be insufficient (Guak et al., 2001).

Table 1 shows the fruit quality of 'Fuji'/M.9 apple treated with the chelate compounds. Fruit weight was the highest (at 232.9 g) in the group treated with 1% Fe-EDTA and the lowest (at 198.8 g and 192.8 g) in the groups treated with 1% Cu-EDTA and 1% Cu-EDTA + 1% Fe-EDTA, respectively. In deciduous fruit trees, nutrients produced in the leaves generally move to the fruit during the maturation period (Loescher et al., 1990). These nutrients have a direct effect on the development of the fruit (Kang and Ko, 1976; Lee and Kang, 1996) since they play a critical role in determining the enlargement of fruit before harvest (Ferree and Palmer, 1982; Roper and Loescher, 1987; Park, 2002). Thus, fruit weight decreased in trees treated with Cu-EDTA and those treated with 1% Cu-EDTA + 1% Fe-EDTA due to the effect of these treatments on photosynthesizing leaves. In these treatments, all fruit cluster leaves, bourse shoot leaves, and true leaves were much more defoliated compared to those in other treatments. On the other hand, the 1% Fe-EDTA treatment resulted in a similar defoliation rate for fruit cluster leaves to that of the other treatments but a lower defoliation rate for true leaves and bourse shoot leaves. This has a direct effect on fruit growth (Choi et al., 2000). Furthermore, the 1% Fe-EDTA treatment resulted in a higher photosynthetic rate for true leaves than in the other treatments. This might have affected fruit weight. Soluble solids content and acidity were not significantly different ( $p > 0.05$ ) among the treatments, consistent with reports of Yim and Lee (1999) and Choi et al. (2000), showing that defoliation did not have an obvious effect on soluble solids content or acidity (Parker et al., 2015).

Anthocyanin contents in peels of harvested fruit were higher in the chelate compound treatments than those in the control, although they were not significantly different among the treatments (Table 2). Fruit coloration, examined using a chromatograph, was the lowest (15.33) for CIE a\* value and the highest (14.63) for CIE b\* value in the control, meaning poor coloring. On the other hand, fruit of the chelate compound-treated groups had better coloration than the control fruit because of the defoliation and improved light interception in the treated groups (Jackson et al., 1971). These results are consistent with several previous reports showing that defoliation can improve fruit coloration (Bae and Lee, 1995; Choi

**Table 1.** Quality of 'Fuji'/M.9 apple fruit harvested after the application of a Cu-EDTA and/or Fe-EDTA foliar spray on 8-year-old 'Fuji'/M.9 apple trees

Treatment <sup>z</sup>	Fruit weight (g)	Firmness (kg·Ø5 mm <sup>-1</sup> )		Soluble solids content (°Brix)	Acidity (%)
		Peel	Flesh		
Control	204.4 bc <sup>y</sup>	3.43 ab	1.30 b	14.3 a	0.344 a
1% Cu-EDTA	198.8 cd	3.35 bc	1.36 b	14.6 a	0.303 a
3% Cu-EDTA	209.2 b	3.48 ab	1.33 b	14.5 a	0.332 a
1% Fe-EDTA	232.9 a	3.15 c	1.39 b	14.8 a	0.382 a
3% Fe-EDTA	212.8 b	3.32 bc	1.35 b	14.6 a	0.367 a
1% Cu-EDTA + 1% Fe-EDTA	192.8 d	3.64 a	1.53 a	14.6 a	0.384 a

<sup>z</sup>Control, water spray.

<sup>y</sup>Mean separation within columns by Duncan's multiple range test at  $p < 0.05$ .

**Table 2.** Fruit coloration and anthocyanin content of 'Fuji'/M.9 apple fruit harvested after the application of a Cu-EDTA and/or Fe-EDTA foliar spray on 8-year-old 'Fuji'/M.9 apple trees

Treatment <sup>z</sup>	CIE value			Anthocyanin content ( $\mu\text{g}\cdot\text{cm}^{-2}$ )
	L*	a*	b*	
Control	48.20 a <sup>y</sup>	15.33 c	14.63 a	0.211 b
1% Cu-EDTA	38.54 c	19.22 b	10.29 c	0.508 a
3% Cu-EDTA	42.09 b	21.01 ab	11.71 b	0.408 a
1% Fe-EDTA	40.78 b	21.34 ab	10.86 bc	0.437 a
3% Fe-EDTA	41.73 b	22.54 a	11.07 bc	0.490 a
1% Cu-EDTA + 1% Fe-EDTA	42.41 b	19.99 b	11.56 b	0.412 a

<sup>z</sup>Control, water spray.

<sup>y</sup>Mean separation within columns by Duncan's multiple range test at  $p < 0.05$ .

et al., 2000; Yim et al., 2000; Nam et al., 2016). Among the treated groups, Fe-EDTA resulted in slightly higher red coloration (CIE a\* value) than Cu-EDTA and Fe-EDTA + Cu-EDTA. This was because Fe-EDTA extensively defoliated fruit cluster leaves, allowing for greater light interception of fruit, whereas it defoliated less bourse shoot leaves and true leaves, which are involved in photosynthesis in the late enlargement period of fruit (Table 2 and Fig. 1). As a result, soluble solids were more highly accumulated in the fruit and used for the production of anthocyanin (Pirie and Mullins, 1976). Regarding the Fe-EDTA concentration, 3% Fe-EDTA induced higher red coloring but lower fruit weight compared to 1% Fe-EDTA. This might be due to the fact that although these two treatments defoliated fruit cluster leaves similarly, 1% Fe-EDTA defoliated nearly half the amount of bourse shoot leaves and true leaves compared to 3% Fe-EDTA.

Taken together, foliar spray of chelate compounds (Cu-EDTA and/or Fe-EDTA) at 30 days before harvest was effective in defoliating fruit cluster leaves, thus affecting light interception of 'Fuji'/M.9 apple trees. However, due to an increased defoliation rate of bourse shoot leaves and true leaves, which are involved in photosynthesis during the late period of fruit growth, Cu-EDTA significantly reduced fruit weight, soluble solids content, and peel coloration compared to Fe-EDTA. A mixed application of 1% Cu-EDTA with 1% Fe-EDTA caused a similar defoliation rate as that of the individual Cu-EDTA treatments and it also significantly reduced fruit weight and peel coloration. The 3% Fe-EDTA treatment resulted in higher peel coloration but lower fruit weight compared to 1% Fe-EDTA. Consequently, the 1% Fe-EDTA treatment was the most effective for improving fruit quality and coloration of 'Fuji'/M.9 apple as it induced higher defoliation of fruit cluster leaves but lower defoliation of bourse shoot leaves and true leaves.

## Literature Cited

- AAES (Aomori Apple Experimental Station) (1985) Effects of defoliation on fruit quality in apples. Ann Rep Aomori Apple Exp Stn, Japan, pp 56-57
- Bae RN, Lee SK (1995) Effects of some treatments on the anthocyanin synthesis and quality during maturation in 'Fuji' apple. J Kor Soc Hortic Sci 36:655-661
- Ben-Yehoshua S, Biggs RH (1970) Effects of iron and copper ions in promotion of selective abscission and ethylene production by citrus fruit and the inactivation of indoleacetic acid. Plant Physiol 45:604-607. doi:10.1104/pp.45.5.604
- Choi SW, Kim JO, Kim KR (2000) Effects of defoliation treatments during maturation on fruit quality of 'Fuji' apples. J Kor Soc Hortic Sci 41:383-386
- Faby R (1988) Using copper chelate for defoliation. Deutsche Baumschule 40:356-357

- Ferree DC, Palmer JW** (1982) Effect of spur defoliation and ringing during bloom on fruiting, fruit mineral level, and net photosynthesis of 'Golden Delicious' apple [varieties]. *J Am Soc Hortic Sci* 107:1182-1186
- Furze ME, Huggett BA, Aubrecht DM, Stolz CD, Carbone MS, Richardson AD** (2019) Whole-tree nonstructural carbohydrate storage and seasonal dynamic in five temperate species. *New Phytol* 221:1466-1477. doi:10.1111/nph.15462
- Guak S, Cheng L, Fuchigami LH** (2001) Foliar urea pretreatment tempers inefficient N recovery resulting from copper chelate (CuEDTA) defoliation of apple nursery plants. *Hortic Sci Biotechnol* 76:35-39. doi:10.1080/14620316.2001.11511323
- Jackson JE, Sharples RO, Palmer JW** (1971) The influence of shade and within-tree position on apple fruit size, color and storage quality. *J Hortic Sci* 46:277-287. doi:10.1080/00221589.1971.11514408
- Kamuro Y, Sakai K** (1995) Convenient synthesis of optically active ABA and plant growth promoting effects of ABA and synthetic intermediates. *Jpn Agrochem Abstr* (spring):140
- Kang SM, Ko KC** (1976) A study on cold hardiness, flowering and fruit bearing in 'Okubo' peach trees (*Prunus persica*) as affected by defoliation. *J Kor Soc Hortic Sci* 17:1-11
- Kim WC, Yun CJ** (1971) Effects of defoliation with chemical spray before harvest on improvement of fruit color in apples. *Ann Rep Hortic Exp Stn*, pp 200-207
- Knight JN** (1983) Chemical defoliation of nursery stock using chelated forms of copper and iron. *J Hortic Sci* 58:471-476. doi:10.1080/00221589.1983.11515145
- Lee YC, Kang SM** (1996) Current and carry-over effects of defoliation on vegetative and fruit growth of Seibel 9119 grapes (*Vitis* spp.). *J Kor Soc Hortic Sci* 37:406-415
- Loescher WH, McCamant T, Keller JD** (1990) Carbohydrate reserves, translocation, and storage in woody plant roots. *HortScience* 25:274-281
- Matsumoto K, Fujita T, Sato S, Chun JP** (2017) Comparison of the effects of early and conventional defoliation on fruit growth, quality and skin color development in 'Fuji' apples. *Hortic Sci Technol* 35:410-417. doi:10.12972/kjhst.20170044
- Millard P** (1996) Ecophysiology of the internal cycling of nitrogen for tree growth. *J Plant Nutr Soil Sci* 159:1-10. doi:10.1002/jpln.1996.3581590102
- Nam JC, Chang EH, Yang H, Cho EK, Jung SM, Hur YY, Koh SW, Choi IM** (2016) Effect of leaf removal on coloration improvement of red variety 'Hongisul' grape. *J Korean Soc Int Agric* 28:231-236. doi:10.12719/KSIA.2016.28.2.231
- Park SJ** (2002) Effect of different degrees of defoliation on fruit quality, reserve accumulation and early growth of young Fuyu persimmon. *Hortic Sci Technol* 20:110-113
- Parker AK, Hofmann RW, van Leeuwen C, McLachlan ARG, Trought MCT** (2015) Manipulating the leaf area to fruit mass ratio alters the synchrony of total soluble solids accumulation and titratable acidity of grape berries. *Aust J Grape Wine Res* 21:266-276. doi:10.1111/ajgw.12132
- Pirie A, Mullins MG** (1976) Changes in anthocyanin and phenolics content of grapevine leaf and fruit tissues treated with sucrose, nitrate, and abscisic acid. *Plant Physiol* 58:486-472. doi:10.1104/pp.58.4.468
- Roper TR, Loescher WH** (1987) Relationship between leaf area per fruit and fruit quality in Bing sweet cherry. *HortScience* 22:1273-1276
- Srisook M, Lim CK, Oh EU, Yi K, Chamidha Kumarihami HMP, Kim SC, Park KS, Song KJ** (2016) Defoliation time influences vine regrowth, off-season flowering, and fruit quality in 'Jecy Gold' kiwifruit vines. *Hortic Environ Biotechnol* 57:219-224. doi:10.1007/s13580-016-0029-5
- Taiz L, Zeiger E** (2002) *Plant physiology*, 3rd Ed. Sinauer Associates, Inc., Publishers, Sunderland, Massachusetts, USA
- Vestrheim S** (1970) Effects of chemical compounds on anthocyanin formation in 'McIntosh' apple skin. *J Am Soc Hortic Sci* 95:712-715
- Yim YJ, Jang JY, Lee HC** (2000) Effect of optically active ABA and its synthetic intermediate STC4771 on defoliation and fruit color in 'Fuji' apple trees. *J Kor Soc Hortic Sci* 41:53-55
- Yim YJ, Lee HC** (1999) Effect of pre-harvest defoliation on fruit color and tree physiology in apples. *J Kor Soc Hortic Sci* 40:209-212